Inter-population variabilities in seed mass and germination of *Panicum turgidum* and *Pennisetum divisum* in the desert of Kuwait

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Abstract: Understanding variability in seed germination among populations is essential for planning an effective germplasm collection for restoration and conservation purposes. The knowledge of germination and dormancy patterns among populations of desert grasses is crucial for determining the potential of the species and populations to be used for restoration and conservation as well as forage production. Variability in seed germination of *Panicum turgidum* Forssk and *Pennisetum divisum* (Gmel.) Henr. in the desert of Kuwait was evaluated in different populations in May 2017. Experiment of seed germination (25 seeds and 4 replicates) was conducted for each population at night/day temperatures of 15°C/20°C and 20°C/30°C under the following light condition: continuous darkness or 12 h/12 h light/dark. Results showed that seed masses of both species strongly varied according to their seed provenances, and both species produced heavier seeds in population with a higher soil electrical conductivity. Seed germination percentage considerably varied between two species, and the variation in *P. turgidum* was greater (17%–49%) than that of *P. divisum* (72%–93%). Germination percentage in *P. turgidum* was greater at high temperature (20°C/30°C) than at low temperature (15°C/20°C). However, temperature regimes had no effect on germination percentage of *P. divisum* seeds. Mean germination time of both species exhibited significant inter-population variability. This result is especially relevant to assure the selection of the best population of each species and the regeneration success of the species. Besides this, inter-population variability also provides valuable information for enhancing our understanding of the mechanisms that regulate seed germination and how they might be related to seed provenance.

Keywords: desert grass; seed germination; inter-population variability; seed provenance; seed dormancy

1 Introduction

Desert is very susceptible to land degradation due to the long-term dry season, low and irregular rainfall, poor soil water holding capacity, mobile surface deposit and sparse vegetation cover (Mganga et al., 2015). Moreover, overgrazing, off-road driving, camping, lack of enforcement for implementing proper resource management policies, industrialization and urban development can cause biodiversity loss and ecosystem degradation due to the degradation of hydrological system

and the loss of soil productivity (Al-Awadhi et al., 2003; Richer, 2008). Planting native plant species for vegetation rehabilitation, which are adapted to local environmental conditions, could be an effective way to restore the degraded desert ecosystems (Bhatt, 2013; Richer et al., 2016).

Variations in altitude, latitude, soil moisture, soil nutrient content, temperature, light and degree of habitat disturbance affect the maternal environmental conditions and consequently influence seed production, dormancy and germination, although the degree to how the maternal environmental conditions affect these phases is frequently species-specific (Baskin and Baskin, 2014; Carón et al., 2014; El-Keblawy et al., 2016, 2017). Therefore, to find out the potential seed resources for vegetation rehabilitation, we should research the inter-population variations in seed production, dormancy and germination from different seed provenances.

Grasses play important roles in animal nutrition and help in land stabilization by soil protection and sand dune fixation in desert (Bhatt et al., 2016) and they could be used for the restoration or rehabilitation in degraded rangelands (Osman et al., 2008). Selection of appropriate species is a key factor that determines the success of environmental restoration especially in desert environment (Phondani et al., 2016).

Panicum turgidum Forssk. and *Pennisetum divisum* (Gmel.) Henr. are drought-resistant C_4 perennial grasses that are widely distributed in the desert of Kuwait (Batanouny, 2002). *P. turgidum* prefers to grow in depressions and shallow wadis with more coarse textured sandy and gravelly soil, whereas *P. divisum* prefers to grow in shallow wadis and runnels in deep sandy soil (Hegazy and Lovett-Doust, 2016). Both species are important sources of fodder for camels and sheep due to their high nutritional values (Asharf, 2006; Rafay et al., 2013). Therefore, they could be the perfect choices for sustainable forage production. In addition, these species could be used to restore and rehabilitate rangelands or to enhance their productivities (Peacock et al., 2003; El-Keblawy, 2013). Both *P. turgidum* and *P. divisum* have also been prioritized for sustainable landscaping because these species have the ability to stabilize the loose soil and therefore they could be used to protect the soil from erosion caused by wind and water (Akhter and Arshad, 2006).

Propagation of any species can only be achieved by selection of superior populations. Therefore, screening the best seed source for certain specific environment is desirable to achieve the maximum productivity (Loha et al., 2009). However, such information is lacking for most of the native desert species including *P. turgidum* and *P. divisum*. Previous studies have shown that seeds of *P. turgidum* and *P. divisum* have physiological dormancy and thus they required after-ripening (Al-Shamsi, 2009; El-Keblawy, 2013). Effects of various environmental factors (i.e., temperature, light and salt stress) and different dormancy regulating hormones have been tested on seed germination of *P. turgidum* and *P. divisum* using seeds from a single population (El-Keblawy, 2004, 2011, 2013; Al-Khateeb, 2006; Taisan, 2010). However, inter-population variability in these species was not evaluated until now. In the present study, we hypothesized that seeds collected from different populations of *P. turgidum* and *P. divisum* with similar climatic conditions will have similar germination behavior. To test this hypothesis, we examined the following questions: (i) is there a difference in seed mass according to the seed provenance? (ii) are there any light and temperature mediated mechanisms affecting germination under laboratory conditions? and (iii) is there a different response of seed germination to light and temperature according to the seed provenance?

2 Materials and methods

2.1 Seed collection and seed mass measurement

The climate is characterized by a long, dry and hot summer, with daytime temperature exceeding 50°C (Annual Statistical Report, 2006) in Kuwait. Mean annual precipitation is less than 114 mm, mostly occurring during winter (Omar et al., 2007). Ripe seeds of *P. turgidum* and *P. divisum* were collected from different populations in the desert of Kuwait in May 2017 (Table 1). Seeds were collected from 25 to 30 randomly chosen plants for both species from each population. Seeds of both species were immediately tested for germination within a week after collection. The seed

mass of each population was determined by weighing 25 seeds with three replicates.

Table 1 Population codes and habitat details of *Panicum turgidum* and *Pennisetum divisum*

Population	Code	GPS coordinates	Associated species
		Panicum turgidum	
KISR (Kuwait Institute for Scientific Research)	KISR	29°09'50"N; 47°41'24"E	Pure stand
Near Julaia	NJ	28°56'10"N; 48°11'21"E	*Heliotropium bacciferum*, *Cenchrus ciliaris* and *Pennisetum divisum*
Sulaibia	SNB	29°06'43"N; 47°24'51"E	*Cyperus conglomeratus*, *Plantago boissieri* and *Calligonum comosum*
Saad Al Abdulla Al Sabah road	SAA	29°24'15"N; 47°41'37"E	*Lasiurus sindicus*, *Stipagrostis plumosa*, *Polycarpae arepens* and *Cyperus conglomeratus*
		Pennisetum divisum	
KISR	KISR	29°07'42"N; 47°43'07"E	Pure stand
Sulaibia	SNB	29°06'44"N; 47°24'52"E	*Plantago boissieri*, *Cyperus conglomeratus* and *Calligonum comosum*
Al Liyah-1	AL1	28°53'19"N; 48°14'27"E	*Deverra triradiata*, *Panicum turgidum* and *Halothamnus iraqensis*
Al Liyah-2	AL2	28°55'38"N; 48°12'07"E	*Salsola imbricata*, *Cyperus conglomeratus* and *Aeluropus lagopoides*
Saad Al Abdulla Al Sabah road	SAA	29°24'05"N; 47°38'24"E	*Cornulaca monacantha*, *Panicum turgidum* and *Haloxylon salicornicum*

Note: Population codes are the same explained in Figures 1, 3 and 4 and Table 2.

2.2 Soil physical and chemical properties

Soil samples were randomly collected from each population at 10 cm soil depth. Soil pH and soil electrical conductivity (EC) were determined by the saturated paw method (AFNOR, 1987). Organic matter (OM) was determined by the partial oxidation method (Walkley and Black, 1934). Furthermore, soil samples were analyzed for total nitrogen (TN), available phosphorus (AP), potassium (K), sodium (Na), calcium (Ca), magnesium (Mg), chloride (Cl) and sulphate (SO_4). TN was determined by the Kjeldahl method, AP was extracted with 0.5 N $NaHCO_3$, Ca and Mg were determined by atomic absorption spectrophotometry, Na and K were determined by flame photometer, Cl was determined by silver nitrate titration and SO_4 were determined by standard gravimetric methods. All of these methods are referenced from Jackson (1973) and Allen et al. (1986).

2.3 Seed germination

For the tested population of each species (4 populations for *P. turgidum* and 5 populations for *P. divisum*; Table 1), germination percentage was evaluated using 25 seeds with four replicates at night/day temperatures of 15°C/20°C and 20°C/30°C under the following light conditions: continuous darkness or 12h/12h light/dark. These temperature regimes were chosen because they are close to the natural climate conditions during February and April in Kuwait (Omar et al., 2007). In the dark treatment, the Petri-dishes were wrapped with two layers of aluminum foil (Parafilm, Sigma-Aldrich, UK). The germination was conducted in 9-cm tight-fitting Petri-dishes containing one disk of filter paper (Whatman No. 1), moistened with distilled water. Seeds were considered to be germinated after the cotyledons appeared. Germinated seeds were counted and removed every alternate day until 28 d. Germinated seeds in the dark treatment were counted after 28 d. A seed was considered to be viable when a cross-section showed that the endosperm was white and appeared healthy. A cut-test was performed to test the embryo status under a binocular microscope that is used to evaluate the viability of un-germinated seeds at the end of germination experiment.

2.4 Data analysis

For these two species, the seed mass was analyzed as a function of seed provenance with a

Kruskal-Wallis test. The germination of both species was analyzed as a function of population (P) and incubation conditions, i.e., light (L) and temperature (T) with generalized linear models. The mean germination time (MGT) of both species was analyzed as a function of P and T with generalized linear models and Gaussian error structures in R version 3.3.2 (R Core Team, 2016). Meanwhile, chi-square tests were performed to make sure that the model fit did not significantly decrease.

3 Results

3.1 Seed mass

The seed masses of both species strongly varied according to their seed provenances. The heavier seeds of *P. turgidum* were produced in population KISR and the lighter seeds in population NJ (chi-square=8.195, *df*=3, *P*=0.042). On the other hand, the heavier seeds of *P. divisum* were produced in population SAA and the lighter in population SNB (chi-square=10.126, *df*=4, *P*=0.038; Fig. 1). Both species produced the heavier seeds in population with a higher soil EC (Fig. 2; Table 2).
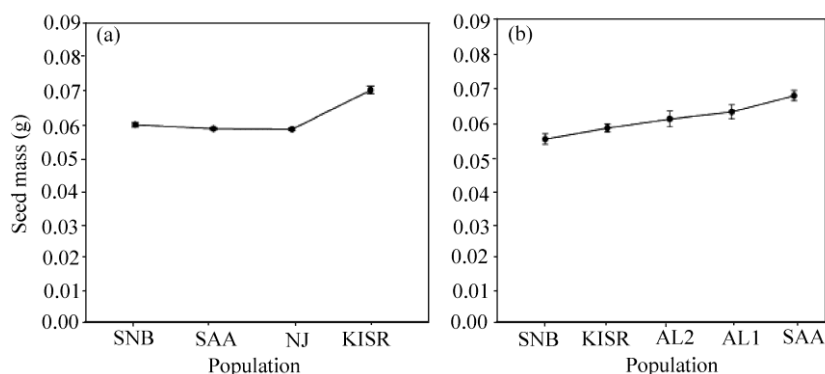

Fig. 1   Seed masses of *P. turgidum* (a) and *P. divisum* (b) in different populations. Bars indicate standard errors.
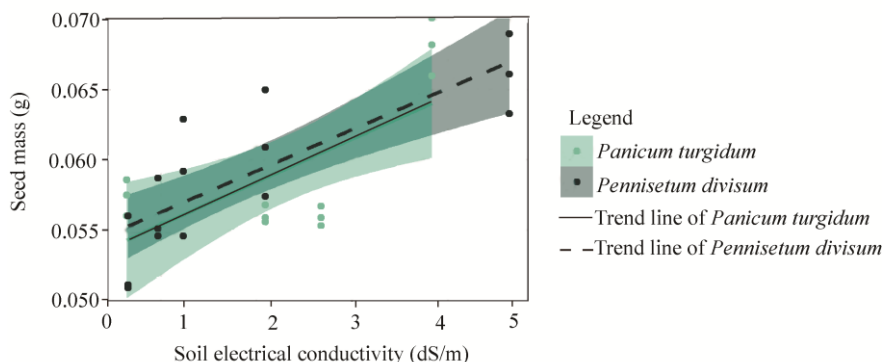

Fig. 2   Seed masses of *P. turgidum* and *P. divisum* along a gradient of soil electrical conductivity

3.2 Germination percentage

Both species showed a strong and significant variation in germination percentage among different seed provenances. Differences of 31.7% and 20.7% were detected between the population with the highest germination and the population with the lowest germination of *P. turgidum* and *P. divisum*, respectively (Fig. 3). The highest germination percentage was recorded in population SAA and the lowest in population KISR for *P. turgidum*, while, for *P. divisum* the highest

Table 2   Soil characteristics of *P. turgidum* and *P. divisum* in different populations

Population	pH	EC (dS/m)	TN (mg/kg)	AP (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	Na (mg/kg)	K (mg/kg)	Cl (mg/kg)	SO$_4$ (mg/kg)	OM (%)
					Panicum turgidum						
KISR	7.7	3.830	360	33.0	141.0	30.6	57.8	35.4	161.3	176.1	0.80
NJ	8.1	2.670	100	2.8	72.1	8.1	42.2	5.5	75.0	148.3	0.29
SNB	8.0	0.640	95	3.0	24.9	2.7	2.0	5.8	5.1	23.6	0.25
SAA	7.6	2.088	110	8.6	81.1	5.7	19.1	15.0	58.5	122.0	0.44
					Pennisetum divisum						
KISR	7.8	0.965	540	12.8	49.5	3.4	17.1	9.1	24.8	64.4	1.16
SNB	7.8	0.656	160	3.8	25.8	0.4	4.9	5.0	8.1	20.1	0.42
AL1	7.6	2.088	110	8.6	81.1	5.7	19.1	15.0	58.5	122.0	0.44
AL2	7.8	1.230	220	11.2	39.4	4.7	15.4	4.8	28.5	68.7	0.92
SAA	7.4	4.640	115	12.2	144.2	21.5	80.9	21.1	63.3	326.0	0.30

Note: EC, electrical conductivity; TN, total nitrogen; AP, available phosphorus; Ca, calcium; Mg, magnesium; Na, sodium; K, potassium; Cl, chloride; SO$_4$, sulphate; OM, organic matter.
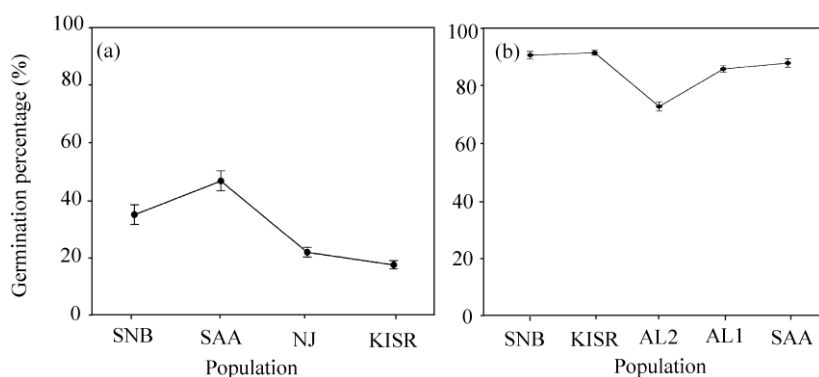

Fig. 3 Germination percentages of *P. turgidum* (a) and *P. divisum* (b) in different populations. Bars indicate standard errors.

germination percentage was found in population KISR and the lowest in population AL2 (Fig. 3). Germination percentage of *P. turgidum* was significantly influenced by temperature (Table 3). Germination percentage of *P. trugidum* was 15.0% higher at high temperature (20°C/30°C) than at low temperature (15°C/20°C; Fig. 4a). On the other hand, germination percentage of *P. divisum* was influenced by light, as slightly higher germination was found under cycle of light and darkness than under complete darkness (Table 3).

Table 3 Effects of population (P), temperature (T) and light (L) and their interactions on seed germinations of *P. turgidum* and *P. divisum*

Treatment	*Panicum turgidum*			*Pennisetum divisum*		
	df	LRT	P	df	LRT	P
P	3	120.631	2.2e-16[***]	4	86.198	<2e-16[***]
T	1	45.902	1.243e-11[***]	1	4.862	0.738[ns]
L	1	0.842	0.359[ns]	1	4.862	0.028[*]
T×L	1	0.012	0.901[ns]	1	0.000	0.994[ns]
P×L	3	2.713	0.438[ns]	4	4.655	0.325[ns]
P×T	3	8.873	0.031[*]	4	9.476	0.049[*]
P×T×L	3	3.315	0.346[ns]	4	0.509	0.973[ns]

Note: df, degree of freedom; LRT, likelihood ratio test; [ns], not significant; [*], significances at $P<0.05$ level; [***], significances at $P<0.001$ level.

Germination percentage of both species was significantly influenced by the interaction between population and temperature (Table 3). For *P. turgidum*, a difference of 45.0% in germination

percentage was found between population SAA and population KISR at high and low temperatures; meanwhile, *P. divisum* showed a difference of 23.5% between population KISR and population AL2 at high and low temperatures (Figs. 4a and b). The difference in *P. turgidum* germination decreased when soil EC increased. Germination percentage of *P. turgidum* population with a high soil EC was almost equal under both temperature regimes (Table 2; Fig. 4). Conversely, no clear pattern related with soil EC was observed in germination percentage of *P. divisum* (Fig. 2). Finally, when comparing the variation in seed mass and germination percentage according to the seed provenance, no clear relationship among these traits was found (Figs. 1 and 3).

3.3 Mean germination time (MGT)

MGT was significantly influenced by the seed provenance (Table 4). MGT of *P. turgidum* in population SNB was 5.76 d, while it was 4.77 d in population NJ. On the other hand, values of MGT of *P. divisum* in population AL2 and population KISR were 2.47 and 2.25 d, respectively (Fig. 4). Additionally, the interaction between population and temperature was significant for both species. MGT of *P. turgidum* was 6.04 d in population SNB at low temperature, while it was 4.14 d in population NJ at high temperature. On the other hand, for *P. divisum* it was 2.49 d in population AL2 at low temperature and 2.33 d in population AL1 at high temperature (Figs. 4c and d).

Table 4 Effects of population (P) and temperature (T) and their interaction on mean germination time of *P. turgidum* and *P. divisum*

Species	Treatment	*df*	Scaled deviation	*P*
Panicum turgidum	P	3	10.343	0.016[*]
	T	1	2.894	0.122[ns]
	P×T	3	11.173	0.011[*]
Pennisetum divisum	P	4	38.522	8.745e-08[***]
	T	1	0.029	0.865[ns]
	P×T	4	11.082	0.026[*]

Note: *df*, degree of freedom; [ns], not significant; [*], significances at $P<0.05$ level; [***], significances at $P<0.001$ level.
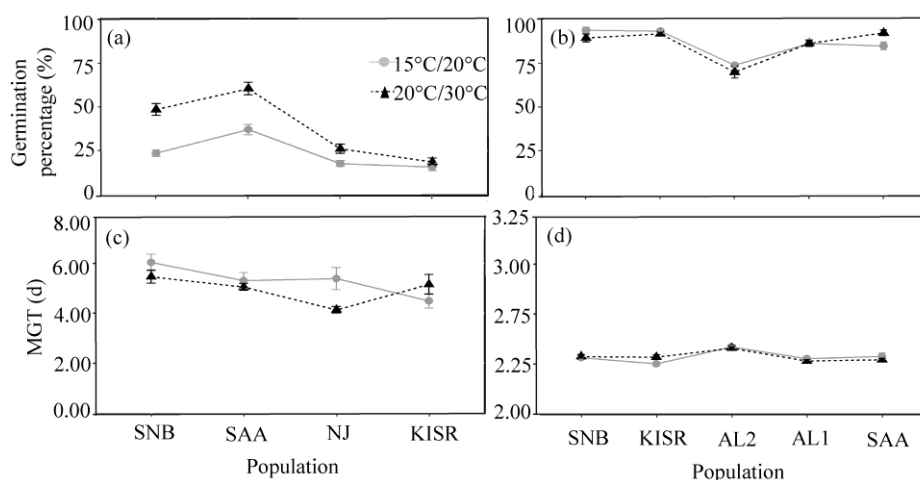

Fig. 4 Germination percentages and mean germination time (MGT) of *P. turgidum* (a and c) and *P. divisum* (b and d) in different populations and temperatures. Bars indicate standard errors.

4 Discussion

Knowledge of variability among populations is essential for planning effective strategies for germplasm collection destined to conservation and restoration (Eriksson, 2014). In the

study, seeds collected from different populations of *P. turgidum* and *P. divisum* showed considerable inter-population variation in seed mass. Difference in seed mass within a species has important ecological consequences because seed mass can affect seed water relation, seed dispersal, germination percentage and seedling establishment (Zhang and Maun, 1990). Maternal environmental conditions such as climatic conditions, soil nutrients and soil moisture content play important roles in determining seed mass (Quesada et al., 1996; Wright et al., 1999; Tremayne and Richards, 2000). Considering that the seeds of *P. turgidum* and *P. divisum* were collected from populations with similar climatic conditions, it is possible that the variation in resource availability among different populations or genetic characteristics of seed provenance could be responsible for the variability in seed mass of both species. Among the studied soil chemical properties, only soil EC shows an apparent positive relationship with seed mass of both species. EC is an indicator of soil salinity and generally, in salt-affected soils, nutrient uptake and use efficiency of plant are low due to this source of stress (Fageria et al., 2011). Therefore, we assumed that the species growing in high saline habitat might allocate more resources to the reproduction and hence the seeds collected from slightly high saline habitat have a higher seed mass. For example, Boot et al. (1986) and Steyn et al. (1996) found that plants can alter the resource allocation to growth and reproduction under environmental stress. In the study, *P. turgidum* seeds that were collected from populations with a high soil EC, germinated equally under both temperature regimes, indicating that seeds collected from similar habitats may have similar temperature requirements during seed germination. The germination percentage strongly varied among populations of *P. turgidum* (17%–49%) and *P. divisum* (72%–93%). These variations in germination percentage among populations are not rare. Previous studies highlighted that seeds from different populations of desert species such as *Anabasis setifera*, *Centaurium somedanum*, *Helianthemum salicifolium* and *Suaeda aegyptiaca* can vary in their dormancies and germinations (Fernández-Pascual et al., 2013; El-Keblawy et al., 2016; Yeşilyurt et al., 2017). Such variations in dormancy and germination among populations have been linked with the ability of species to persist under variable environmental conditions by producing seeds with different dormancy status, thus reducing the risk of synchronizing germination and increasing the fitness for germination under various environmental conditions (Cochrane et al., 2015; Santo et al., 2015). Previous studies reported that *P. turgidum* and *P. divisum* exhibit different levels of physiological dormancy (Al-Shamsi, 2009; El-Keblawy, 2013). Considering the strong variation in germination percentage among populations reported here, we suggest that no conclusions regarding germination of these species could be drawn. Moreover, the seed provenance needs to be carefully considered before interpreting the germination behaviour in wild species because this information could help in understanding the survival strategies of species growing under unpredictable environmental conditions (Gutterman, 1994; Kigel, 1995).

In the present study, germination percentage of both species varied according to the seed provenance and the temperature. *P. turgidum* seeds showed a better germination at high temperature (20°C/30°C) irrespective of seed provenance. These results are in contradiction with Al-Khateeb (2006) and Al-Shamsi (2009), who reported that *P. turgidum* germinated well in a wide range of temperatures. In addition, light requirement for optimal germination of *P. turgidum* appeared to vary in each population, implying the inter-population sensitivity to light.

For *P. turgidum*, complete darkness seems to be more effective in enhancing germination at high temperature, which might be an adaptive strategy for desert conditions. If the seeds are remained on the surface layer, they would face strong light, extreme temperatures and drought due to the high evaporation at surface layer, and these factors could, cumulatively, inhibit seedling establishment. Seeds of *P. divisum* germinated well in both light and dark, indicating that light is not an obligatory requirement for germination. Furthermore, the ability of *P. divisum* to germinate in dark could be related to its adaptability to sandy habitat where sand burial often occurs. However, if they are buried too deep in the soil, they might be negatively impacted because they might spend all their reserves to reach the soil surface. Understanding the ideal conditions for seed germination, including temperature, is essential for determining the timing of seed sowing for vegetation restoration (Baskin and Baskin, 2014; Dürr et al., 2015). Likewise,

light requirements for germination could provide indications about the capacity of the species to form soil seed banks and also provide information about the appropriate sowing method (Jones et al., 2004; Bhatt and Pérez-García, 2016).

MGT of both species varied according to the seed provenance. Additionally, MGT for different populations of both *P. turgidum* and *P. divisum* showed variations at different temperatures. This result might be important for these species' regeneration in the field because seeds from certain provenance might exhibit faster germination in the earlier season by taking advantage of earlier rain events (January–February), whereas seeds of other populations may germinate in the later season (during March–April). Seed mass did not have any effect on germination percentage and MGT of both two species. These findings are in agreement with Baskin and Baskin (2014), who reported that germination rate and percentage could increase, decrease or remain unaffected by the difference in seed size.

Inter-population variability in seed mass, germination percentage and MGT has been correlated with the differences in environmental conditions, biotic and abiotic factors, as well as genetic factors (Bewley and Black, 1994; Li and Foley, 1997; Baskin and Baskin, 2014). Although different populations of *P. turgidum* and *P. divisum* experience similar climatic conditions during seed production, these populations might differ in terms of herbivory pressure, inter- and intra-specific competition with respect to soil nutrients and water holding capacity that could be responsible for the observed variation in seed mass and seed germination percentage among populations of both species. However, we cannot confirm that there were genetic differences among populations in germination because our study only evaluates the response to seed provenance.

The variability in germination behavior among populations of *P. turgidum* and *P. divisum* might be advantageous under harsh desert environments, which could favor the establishment of seedlings. Therefore, we suggest that this high variation among seed provenances must be adequately managed and could be useful in identifying the best seed sources for vegetation restoration.

5   Conclusions

This study indicates that *P. turgidum* seeds could germinate from March to early April, the period when the monthly average temperatures ranged from 18°C to 34°C in Kuwait (data not shown). However, seeds of *P. divisum* could be sown at any time when there is a possibility of rain. Our study also confirms that inter-population variation in seed traits was found in both species and could be useful for vegetation restoration and management. However, field studies will be necessary to resolve the adaptive effects of phenotypes of species.

Acknowledgements

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